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**Dental mesenchymal stem cells encapsulated in an alginate hydrogel co-delivery microencapsulation system for cartilage regeneration.**

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**Public Summary:**

Our results highlight the vital role played by the microenvironment, as well as value of presenting inductive signals for viability and differentiation of mesenchymal stem cells (MSCs).

**Scientific Abstract:**

Dental-derived mesenchymal stem cells (MSCs) are promising candidates for cartilage regeneration, with a high capacity for chondrogenic differentiation. This property helps make dental MSCs an advantageous therapeutic option compared to current treatment modalities. The MSC delivery vehicle is the principal determinant for the success of MSC-mediated cartilage regeneration therapies. The objectives of this study were to: (1) develop a novel co-delivery system based on TGF-beta1 loaded RGD-coupled alginate microspheres encapsulating periodontal ligament stem cells (PDLSCs) or gingival mesenchymal stem cells (GMSCs); and (2) investigate dental MSC viability and chondrogenic differentiation in alginate microspheres. The results revealed the sustained release of TGF-beta1 from the alginate microspheres. After 4 weeks of chondrogenic differentiation in vitro, PDLSCs and GMSCs as well as human bone marrow mesenchymal stem cells (hBMMSCs) (as positive control) revealed chondrogenic gene expression markers (Col II and Sox-9) via qPCR, as well as matrix positively stained by Toluidine Blue and Safranin-O. In animal studies, ectopic cartilage tissue regeneration was observed inside and around the transplanted microspheres, confirmed by histochemical and immunofluorescent staining. Interestingly, PDLSCs showed more chondrogenesis than GMSCs and hBMMSCs ( $p < 0.05$ ). Taken together, these results suggest that RGD-modified alginate microencapsulating dental MSCs make a promising candidate for cartilage regeneration. Our results highlight the vital role played by the microenvironment, as well as value of presenting inductive signals for viability and differentiation of MSCs.

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